Comparison of Methane Production from Bench- and Sub Pilot-Scale Anaerobic Digesters

S. T. Sell, R. T. Burns, L. B. Moody, D. R. Raman

Abstract. *Design and construction of full-scale anaerobic digesters that co-digest manure with other substrates, such as food processing wastes, is challenging because of the large number of potential mixtures that can be fed to the digester. In this work we examine the relationship between results from bench-scale methods such as biochemical methane potential assays (BMPs) and sub pilot-scale reactors. The baseline feedstock for this study was beef manure from concrete feedlot pens (open and covered) in eastern Iowa. Additional co-digestion substrates tested were short-fiber cardboard, corn processing wastewater, enzyme processing wastewater and lagoon liquid. Substrates were characterized for total solids (TS), volatile solids (VS), chemical oxygen demand (COD), pH, alkalinity, and ammonia, after which BMPs were conducted on all substrates. Based on the BMP and anaerobic toxicity assay (ATA) results, a mixture was created and evaluated using BMPs and tested in 100-L sub pilot-scale reactors. This study showed that results from BMPs of feedstock co-digestion mixtures accurately estimated the range of methane produced from three 100-L, plug flow reactors.*

Keywords. *Anaerobic digestion, Biochemical Methane Potential (BMP), Co-digestion, Reactor.*

Co-digestion of animal manure with industrial wastewaters or other sources of biodegradable materials for increased energy production is becoming popular in the United States (Braun and Wellinger, 2003). However, full-scale anaerobic digestion (AD) reliability has been low due to system design and management challenges (USDA-NRCS, 2007). Design and construction of a full-scale anaerobic digester should be first validated by less expensive, smaller scale procedures that characterize hydraulic retention time (HRT), organic loading rate (OLR), and methane yield (Wilkie et al., 2004). The ideal process begins with laboratory characterization of potential substrates, and then uses biochemical methane potential assays (BMPs) and anaerobic toxicity assays (ATAs) to examine potential mixtures of substrates (Owen et al., 1979).

The BMP is a powerful method of establishing baseline performance data for AD (Speece, 1996; Bishop et al., 2009). While BMPs provide information regarding the methane production of a substrate, they are typically highly diluted and may mask potential substrate toxicity (Moody et al., 2011a). The ATA was developed to evaluate a substrate's ability to inhibit methane production and therefore determine its potential toxicity. Although critical to early stage design, BMP and ATA results may be misleading when applied directly to full-scale operation due to their lack of information addressing HRT, substrate interaction, and continuous organic loading. However, there have been few publications addressing a proper procedure for AD scale up from substrate identification to full-scale operation. The objective of this study was to analyze the ability of the BMP method to predict methane production of larger scale anaerobic digestion processes. To do so, this article reports on the performance of individual substrates and a substrate mixture in BMPs and 100-L, plug-flow sub pilot-scale anaerobic digesters.

Submitted for review in January 2011 as manuscript number SE 9009; approved for publication by the Structures & Environment Division of ASABE in May 2011.

The authors are **Steven T. Sell, ASABE Member,** Graduate Research Assistant, Iowa State University, Ames, Iowa; **Robert T. Burns, ASABE Member,** Assistant Dean, University of Tennessee, Knoxville, Tennessee; **Lara B. Moody, ASABE Member,** Director of Stewardship Programs, The Fertilizer Institute, Washington D.C; and **D. Raj Raman,** Associate Professor, Department of Agricultural and Biosystems Engineering, Iowa State University, Ames, Iowa. **Corresponding author:** Steven T. Sell, 3155 NSRIC, Iowa State University, Ames, IA 50011; phone: 515-294-3153; e-mail: sellsteve@gmail.com.

Materials and Methods

Substrates

Manure was obtained directly from confined concrete finishing beef cattle feedlot pens (open and covered) in eastern Iowa, from a facility where corn stover was the primary bedding material. The diet consisted primarily of corn, distiller's grain, and gluten. At the time of collection, the manure's age was 2 to 3 d, and the manure was selected from areas with minimal bedding mixed in. A wet mill corn processing wastewater and crude glycerin from a soybean and animal lard biodiesel manufacturing facility were collected within 1 d of delivery to the farm. Cardboard fibers too short for production for a cardboard box manufacturing facility were collected within 5 d of delivery to the farm. Lagoon liquid was collected directly from the on-farm beef manure and separated digester effluent lagoon using a dipper on the side opposite to the influent pipe for maximum lagoon treatment effects. All samples were collected in 20-L buckets, stored at 4°C, and were analyzed within one week of collection. These substrates were selected out of a list of multiple substrates described by Sell et al. (2010). Selection was based on material availability and on performance in BMPs and ATAs. Industrial wastewaters of choice were not in sufficient quantity to provide all dilution requirements;

  ISSN 0883-8542

therefore, on-site water reuse became essential. Since bedding materials were a portion of the manure, they were not considered as a standalone substrate. Some items are not discussed in this article since they were eliminated from mixture selection such as food scraps and soybean processing wastewater. Food scraps were available in limited amounts on an irregular basis and were eliminated on that basis. The low COD value and long trucking distance of the soybean processing wastewater caused its elimination, while the enzyme production wastewater was eliminated due to its toxicity. The corn processing wastewater pH was observed to drop rapidly upon sitting, possibly hindering AD. However, the facility producing the corn processing wastewater was willing to adjust pH prior to delivery. Experiments were run to explore how mixing with manure would buffer this change. If the corn processing wastewater were adjusted to an initial pH of 8.5 with NaOH, a pH above 6.5 could be held for at least one week with a 10/90 wastewater/manure mixture. The mixture was designed from these substrates to meet criteria including the use of all available manure, keeping total solids below 15% to facilitate pumping, maintaining pH between 6.5 and 8.2 for microbial ecology, providing high COD concentrations to maximize methane production, and with limited ammonia levels to avoid toxicity (Speece, 1996).

Analytical Methods

Substrates and mixtures were characterized for total solids (TS), volatile solids (VS), ammonia, alkalinity, and pH by the Iowa State University Agricultural Waste Management Laboratory. The TS and VS concentrations were measured using standard methods 2540 B and 2540 E, respectively (Standard Methods, 1995). The pH measurements were taken with an Accumet Basic AB15 Plus pH meter and Accumet 13-620-285 pH probe (Fisher Scientific, Pittsburgh, Pa.). The chemical oxygen demand (COD) values were measured using Hach DR/890 Colorimeter Procedures Manual, Method 8000 (Loveland, Colo.) and vials for COD 0-1500 ppm. Ammonia concentrations were measured using standard methods 4500-NH_3-B Preliminary Distillation Step and 4500-NH_3-C Titrimetric Method with 0.1-N HCl as the titrant instead of sulfuric acid (Standard Methods, 1995). Alkalinity was measured using standard methods 2320 B with 0.1-N HCl as the titrant (Standard Methods, 1995). A BMP assay was performed in triplicate for each of the individual substrates and mixtures using a modified version of the International Standard ISO 11734 (1995) per Moody et al. (2011b).

Laboratory TS, VS, and COD results were used to calculate the sample size needed for a 250-mL BMP assay serum bottle (Wheaton Science Products; 250 mL Btl, Serum, Type I Clr, Grad; Millville, N.J.). Sample sizes were calculated with a target of 125-mL CH_4 produced during a 30-day period, assuming 70% of COD converted to CH_4, and 395-mL CH_4/g COD reduced (Speece, 1996). This approach yielded average daily biogas volumes that were in a readily measurable range. The BMP reactors were seeded with an inoculum from a 60-L, mesophilic (35°C), continuously stirred anaerobic reactor that was fed a mixture of high-protein dog food and basal medium (Moody et al., 2011b). The BMP reactors were also seeded with basal medium containing supplemental inorganic nutrients and alkalinity (Speece, 1996). Inoculum was added for a 2:1 mass ratio between substrate and inoculum VS. The amounts of each constituent are shown in table 1 where the total volume was 200 mL.

Table 1. Constituent breakdown for individual and mixture BMPs.

BMP	Substrate Amount	Inoculum (mL)	Basal Medium (mL)
Corn processing wastewater	9 mL	68	123
Short-fiber cardboard waste	1.8 g	132.2	~66
Enzyme processing wastewater	2.7 mL	57.8	139.5
Lagoon liquid	20 mL	17	163
Raw manure	2.8 mL	44.7	152.5
Mixture[a] sample taken at sub-pilot startup	5.5 mL	85	109.5
Mixture[a] sample taken three HRTs into sub-pilot operation	7 mL	100	93

[a] Mixture was composed of (22% raw manure, 14% short-fiber cardboard waste, 16% corn processing wastewater, and 48% lagoon liquid).

Assay bottles were purged with 70% nitrogen and 30% carbon dioxide gas at ~0.5 L min^{-1} for 5 min. Bottles were then capped with septa that were secured with plastic zip ties and incubated at 35°C on an orbital shaker at 150 rpm. Biogas production was measured daily by inserting a glass, gas-tight syringe (Micro-Mate Interchangeable Hypodermic Syringe 50cc Lock Tip, Popper & Sons, Inc., New Hyde Park, N.Y.) into the septum and allowing the biogas pressure to displace the wetted barrel of the syringe. The volume was recorded, and the biogas was injected into an infrared gas analyzer (NDIR-CH_4 Gasanalyzer, University Kiel, Germany) to obtain the methane content (Bishop et al., 2009). A blank that included the inoculum source but no substrate was run so that each BMP could be corrected for the methane created by the inoculum source.

The ATA methodology used at the Iowa State University Agricultural Waste Management Laboratory (ISU AWML) was a modified version of the method performed by Owen et al. (1979) and the International Standard ISO 13641-1 (2003) per Moody et al. (2011a). Aliquots of anaerobic inoculum and an easily degraded standard feedstock were assayed alone (for a fed control) and in combination with a range of eight potential toxicant inclusion rates. The inoculum source was the same as noted in the BMP method. Once materials were combined in the serum bottles, each bottle was purged with a 70% nitrogen and 30% carbon dioxide gas at ~0.5 L min^{-1} for 5 min. Bottles were then capped with septa and zip tied, and incubated at 35°C on an orbital shaker at 150 rpm. Biogas production was measured every 24 h over for up to 5 d or until gas production ceased by inserting a glass syringe into the septum and allowing the biogas pressure to displace the wetted barrel of the syringe. The volume was recorded, and the biogas was injected into an infrared gas analyzer (NDIR-CH_4 Gasanalyzer, University Kiel, Germany) to obtain the methane content (Bishop et al., 2009). Results were used to calculate the percent inhibition of methane production for each substrate inclusion rate. Results are reported on a cumulative methane production over a 5-d period or until methane production has ceased as well as on an inclusion verse inhibition basis. In the inclusion verse inhibition display a negative inhibition percentage indicates that a substrate is non-toxic and a positive inhibition indicates signs of toxicity.

Sub Pilot-Scale Reactors

Sub pilot-scale anaerobic digestion reactors were constructed out of 19.05-mm thick high density polyethylene (HDPE) piping with an inside diameter of 28.45 cm. The HDPE pipes were cut to a length of 2.59 m and circular HDPE flanges were extrusion welded on the ends to create the digester chamber. Schedule 80 polyvinyl chloride (PVC) fittings were attached as shown in figure 1.

Self-regulating heater cable (Nelson Heat Trace, HLT15-J, Tulsa, Okla.) was wrapped around the exterior of each digestion tube and connected to a 120-V wall outlet. Plastic bubble wrap insulation with a foil backing was wrapped around the pipe to reduce heat losses from the reactor. Two type-T thermocouples (Omega Engineering, Inc., EXTT-T-20, Stamford, Conn.) were placed in the reactor at the axial center, one at the radial cross-sectional center of the pipe and the other about 50.8 mm from the internal surface so that both would be submerged in the digestate. The temperature was collected and managed using LabView software (National Instruments Corporation, LabView Version 7.1, Austin, Tex.) through personal measurement devices (Measurement Computing Corporation, USB-1208LS, USB-TC, Norton, Mass.) connected to a PC. The program was set up in a manner to control the temperature of each reactor at 35°C. A 6.35-mm gas port was installed on top of the pipe at the axial center of the digester body and was connected to an inverted tipping-bucket gas meter submerged in water. Each sub pilot-scale digester had a calibrated tipping-bucket gas meter that recorded gas production amounts using a magnetic reed switch (Digi-Key Corporation, 59065-010-ND, 57065-000-ND, Thief River Falls, Minn.) via the LabView program. Methane content was determined using 1-L Tedlar bag samples that were measured using an infrared gas analyzer (NDIR-CH_4 Gasanalyzer, University Kiel, Germany). Each digester was started using 100 L of 50/50 water manure slurry that was allowed to reach 35°C for 1 week. Digester 1 was started approximately 3 HRTs prior to digesters 2 and 3 in order to troubleshoot any operation problems before initiation of data collection. Manure was then added following a 21-d HRT until stable gas production was reached. The feedstock was then switched to the mixture and was manually fed in a semi-batch mode (17 L twice per week) that maintained the 21-d HRT.

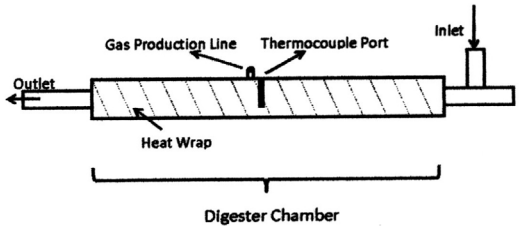

Figure 1. Diagram of sub pilot-scale 100-L, plug-flow anaerobic digester. Flow enters at stand pipe and exits through other side. Heat trace is wrapped around each reactor and covered with plastic insulation with a foil backing. Not shown is continuous temperature control via a PC running LabView and continuous biogas monitoring via inverted tipping-bucket gas meters.

Results and Discussion

Individual substrate characteristics results are shown in table 2. Liquid samples were generally consistent, while solid materials had high variations in some measured variables from week to week (e.g., 15% to 30% TS in manure samples). Subsample results listed in table 2 reflect an average of stockpiles, and we used representative samples for the BMP assays.

It is important to note that the enzyme processing wastewater appeared to be an ideal dilution liquid based on its BMP results; however, an ATA revealed that even at very low inclusion rates, the wastewater was toxic to the anaerobic consortia. The ATA was determined by comparing methane production from a series of enzyme processing wastewater inclusion rates to a known degradable feedstock (Moody et al., 2011a). It was speculated that the toxicity was due to high ammonia concentrations; therefore, the substrate was dropped as a mixture candidate. A comparison of the selected substrate mixture characteristics is shown in table 3 and both the average observed values and the predicted values based on a weighted average of the individual component analyses are listed. The observed mixture characteristics represent an average based on influent samples collected weekly for 15 weeks. The differences in the observed and predicted values likely reflect the variable solids in raw manure and short-fiber cardboard waste. However, the COD/VS ratios observed remained very close to the predicted values (Additional BMP results for these mixed wastes are available in Sell et al., 2010.).

A BMP test is a predictive measurement of anaerobic digestion methane production and was not considered to be a multiple long-term operation test for field application; therefore, we chose to mimic the setting for which a BMP would be used by performing two sets of mixture BMPs in triplicate. Mixture BMPs were conducted prior to starting the sub pilot-scale reactors, and on the 3rd HRT or slightly past the half-way point in the process. The start-up BMP set was used to provide estimates of the methane production;

Table 2. Characteristics of selected substrates.

Substrate	TS (%)	VS (%)	pH	COD[a] (mg/g or mg/L)	Ammonia (mg NH_3-N/L)	Alkalinity (mg $CaCO_3$/L)	BMP (mL CH_4/g VS)
Off-site co-substrates							
Corn processing wastewater	8.3(0.05)	7.6 (0.05)	4.02	107,600(4,500)[a]	260(10)	0	266(42)
Short-fiber cardboard waste	49.0(0.32)	39.4(0.19)	–	406(61)	400(80)	7,900(370)	208(16)
Enzyme processing wastewater	12.8(0.04)	11.3(0.04)	5.05	162,500(9,200)[a]	3,330(200)	3,190(40)	284(10)
On-site materials							
Lagoon liquid	1.3(0.04)	0.9(0.03)	7.06	22,500(1,250)[a]	2,900(200)	8,560(400)	356(33)
Raw manure	17.0(0.50)	14.0(0.81)	6.60	156(28)	1,980(280)	6,000(330)	101(19)

[a] COD reported in mg/L. Values in parenthesis are standard deviations.

Table 3. Characteristics of the substrate mixture, showing actual measurement and the predicted values based a weighted average of the individual components.[a]

Estimation Type	TS (%)[b]	VS (%)	pH	COD (mg/L)	Ammonia (mg NH_3-N/L)	BMP (mL CH_4/g VS)
Predicted based on individual analyses	11.8	9.6	–	110,600	1,910	202[c]
Average of actual measurements	9.2(2.05)	7.2(1.52)	6.50	80,200(4,930)	2,150(100)	178 (6)[d] 124 (6)[e]

[a] This mixture was 22% raw manure, 14% short-fiber cardboard waste, 16% corn processing wastewater, and 48% lagoon liquid by volume.
[b] Values in parenthesis are standard deviations.
[c] Predicted on a mass fraction basis from individual results.
[d] Original BMP performed during sub pilot-scale startup.
[e] BMP performed during 3rd HRT of sub pilot-scale operation.

however, it did not have a range large enough to capture all methane production levels of the sub pilot-scale reactors. Therefore, the second BMP set was performed to extend the range and better estimate the methane production. The highest BMP value found, which occurred in the start-up BMP set, and the lowest BMP value, which occurred in the second BMP set, were multiplied by the average VS loading rate to estimate the maximum and minimum BMP-predicted, daily gas production lines for the sub pilot-scale reactors. These maximum and minimum values are indicated as dashed horizontal lines on figure 2. There was no rational in selection of the time in which the second BMP set was performed, other than it was necessary since the start-up BMP, by itself, did not predict daily methane production rates of the sub pilot-scale digesters for the entire duration of the study. The observed methane flows from the sub pilot-scale anaerobic digesters are displayed in figure 2. Data recording began (0.0 HRT) when the feed was switched from only manure to the mixture discussed in table 3. Data between 1.43 and 1.62 HRTs were omitted due to their loss during a power outage. The power outage also caused a failure of temperature control, and appears to have led to depressed gas production in the time immediately after the outage (fig. 2).

In figure 2, the early methane production is above the predicted maximum value (based on the BMPs). This is likely due to degradation of remnants of the inoculum during this time. Digester performance subsequently became more stable in all three reactors, but variations in methane production continued, most likely due to changes in feedstock. The results indicate that mixture BMPs were reasonable predictors of a methane production range for three 100-L plug flow anaerobic digesters. The results also show that BMPs are a snapshot of a real waste, and that temporal variations in the waste can lead to variations in the performance of larger reactors. But equally, the results show that "identical" reactors fed the same waste can have significant variations in gas production. This reflects a combination of the inherent variability of these biological processes and the difficulties in achieving identical conditions in sub pilot-scale reactors fed on mixed wastes.

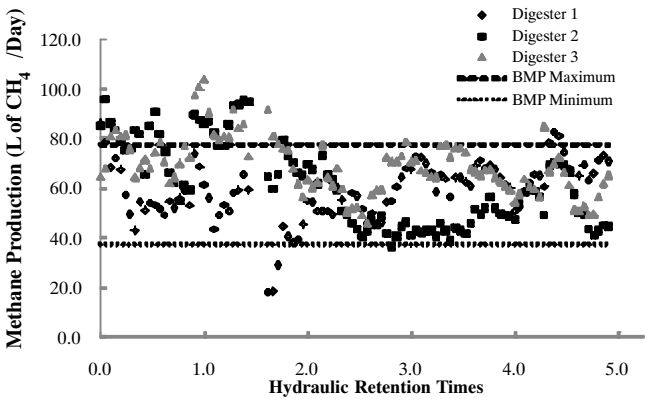

Figure 2. Daily methane production for each sub pilot-scale digester, in comparison to BMP defined gas production values of the mixture. One hydraulic retention time is equivalent to 21 days.

Conclusion

Determining the best mixture for full-scale anaerobic co-digesters is challenging. This work examined the relationship between results from bench-scale methods such as biochemical methane potential assays (BMPs) and sub pilot-scale reactors. Substrates were characterized for multiple parameters and BMPs were conducted on all substrates. A mixture was designed based on BMP and ATA results, and this mixture was tested in 100-L sub pilot-scale reactors. The BMP maximum and minimum were found to be valid boundaries for the sub-pilot scale ADs after 2 HRTs. Bench-scale methods were helpful in determining larger scale performance while the sub pilot-scale testing allows materials handling issues (e.g., floating solids, clogging) to be identified, and provides more robust data for an economic analysis based on realistic biogas production rates.

References

Bishop, G. R., R. T. Burns, T. A. Shepherd, L. B. Moody, C. Gooch, R. Spajić, and J. Pronto. 2009. Evaluation of laboratory biochemical methane potentials as a predictor of anaerobic dairy manure digester biogas and methane production. ASABE Paper No. 096769. St. Joseph, Mich.: ASABE. Available at: asae.frymulti.com/azdez.asp?search=1&JID=5&AID=27205&CID=reno2009&v=&i=&T=2.

Braun, R., and A. Wellinger. 2003. Potential of co-digestion. IEA Bioenergy. Task 37 – Energy from biogas and landfill gas. Available at: www.iea-biogas.net/_download/publi-task37/Potential%20of%20Codigestion%20short%20Brosch221203.pdf.

ISO. 1995. Water quality – evaluation of the "ultimate" anaerobic biodegradability of organic compounds in digested sludge – method by measurement of biogas production. International Standard Organization 11734. Genève, Switzerland: ISO.

ISO. 2003. Water quality – determination of inhibition of gas production of anaerobic bacteria – Part 1: General test. International Standard Organization 13641-1. Genève, Switzerland: ISO.

Moody, L. B., R. T. Burns, S. T. Sell, and G. Bishop. 2011a. Using anaerobic toxicity assays to aid in co-substrate selection for co-digestion. *Applied Eng. in Agric.* 27(3): 441-447.

Moody, L. B., R. T. Burns, G. Bishop, S. Sell, and R. Spajić. 2011b. Using biochemical methane potential assays to aid in co-substrate selection for co-digestion. *Applied Eng. in Agric.* 27(3): 433-439.

Owen, W. D., D. Stuckey, J. Healy Jr., L. Young, and P. McCarty. 1979. Bioassay for monitoring biochemical methane potential and anaerobic toxicity. *Water Res.* 13: 485-492.

Sell, S. T., R. T. Burns, D. R. Raman, and L. B. Moody. 2010. Approaches for selecting anaerobic digestion co-substrates for a full-scale beef manure digester using biochemical methane potentials and anaerobic toxicity assays. In *Proc. 2010 Intl. Symp. On Air Quality and Waste Mgmt. for Agric.* St. Joseph, Mich.: ASABE. Available at: asae.frymulti.com/azdez.asp?search=1&JID=1&AID=32679&CID=isaq2010&v=&i=&T=2.

Speece, R. 1996. *Anaerobic Biotechnology for Industrial Wastewaters*. Nashville, Tenn.: Archae Press.

Standard Methods for the Examination of Water and Wastewater. 1995. 19th ed. Washington, D.C.: American Public Health Association.

USDA-NRCS. 2007. An analysis of energy production costs from anaerobic digestion systems on U.S. livestock production facilities. Tech. Note No. 1. Washington, D.C.: USDA-NRCS.

Wilkie, A. C., P. H. Smith, and F. M. Bordeaux. 2004. An economical bioreactor for evaluating biogas potential of particulate biomass. *Bioresource Tech.* 92: 103-109.